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Blue Mold Decay of Delicious Apples in Relation to Handling Practices

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IMPORTANCE OF DECAY AND CONTRIBUTING FACTORS

Delicious apples now constitute a large proportion of the commercial crop in Washington, and many of them-about a third in 1940-are marketed after February 1. This fact, together with the increased awareness among shippers that high quality in the Delicious variety can be maintained through a long storage season only by prompt cooling to 30° to 32° F., has necessitated changes in some of the older handling practices. Under present conditions Delicious apples are harvested faster than they can be packed, with the result that much of the fruit must be held either in the orchard or the warehouse at high temperature or in cold storage prior to washing and packing. Such holding in cold storage has the decided advantage of maintaining dessert quality, but some plant operators contend that passing cold, turgid fruit through heated washing solutions results in increased injury and subsequent decay.

The extension of the storage and marketing season for Delicious apples has increased the hazard of fungus decay, most of which is

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caused by blue mold (*Penicillium expansum* Lk. emend. Thom), despite the fact that mechanical injuries such as stem punctures and box cuts have been greatly reduced in commercially packed fruit. One of the reasons that serious decay losses continue is that lenticels and other microscopic breaks in the skin frequently serve as avenues of infection (fig. 1). Some of the factors related to the infection of



apples at lenticels have been investigated by other workers, but little has been known of the effect of various washing and storing practices on the susceptibility of the fruit to this type of fungus invasion.

The studies reported in this circular were conducted during the period from 1938 to 1943 to determine the effects of certain handling practices on the development of blue mold decay in Delicious apples. Of primary concern was

FIGURE 1.—Lenticel decay caused by blue mold.

the problem whether cold storage prior to washing and packing results in increased washing injury and decay. Other factors investigated were the severity of the washing treatment, the drying of fruit prior to packing, and the population of blue mold spores in the washing solutions and rinse tank. In addition, a histological study of the skin of the fruit was made to clarify some of the questions relative to the penetration of lenticels and other microscopic openings by blue mold.

In general during the present study more open lenticels and greater amounts of lenticel decay were found on washed than on unwashed fruit when both were dipped in a suspension of blue mold spores before they were packed. Unwashed fruit dipped in a spore suspension developed more lenticel infection than comparable washed fruit packed without being dipped. Thus, it is evident that unwashed fruit with a high natural spore population might develop more decay than fruit washed under sanitary conditions and rinsed with plenty of clean water, even though the washed fruit might be more susceptible to decay.

A delay of 3 days in a nonrefrigerated warehouse decreased the susceptibility of the fruit to washing injury and blue mold infection. Although such a delay cannot be recommended, it should be borne in mind that increased decay and washing injury might occur if immediate washing and packing were put into general practice without concurrent precautions to reduce the spore population and the severity of the washing treatment.

No justification was found for the commonly held belief that washing and packing apples after they have been held for several weeks at 32° F. make them more subject to washing injury and decay than

immediate washing. Cold storage for 2, 6, or 10 weeks prior to washing resulted in increased resistance to washing injury, the increases in general being proportional to the length of the storage period. When fruit was held for 6 weeks prior to washing, its resistance to infection was consistently increased.

Fruit packed while wet developed no more decay than that dried before packing.

Passing apples through a heavily contaminated washing solution followed by a relatively clean rinse caused only a slight increase in the amount of decay, whereas subjecting the unwashed fruit to a special rinse containing a high population of blue mold spores resulted in a greatly increased number of infections.

Lenticels and washing injuries constituted the principal courts of infection in apples subjected to a severe dual-process washing treatment. Milder treatments resulted in an approximately equal distribution of the infections between lenticels and mechanical injuries; in unwashed fruit mechanical injuries predominated in promoting invasion.

Susceptibility to blue mold infection varied in fruit from different orchards and in that from the same orchard during successive seasons.

No clear-cut relation could be found between the number of open lenticels, as determined by dye penetration, and the number of infections that developed at lenticels. However, a severe washing process was found to increase both the number of open lenticels and the number of lenticel infections.

In addition to true lenticels, many other minute lenticellike openings that served as avenues of infection were found in the skin of the fruit. Some of these resulted from the washing process, but the origin of others was obscure.

The importance of careful handling in the control of blue mold decay is emphasized by the fact that many of the lenticel infections occurred in bruised areas.

Although a number of fungi were isolated from lesions originating at mechanical injuries, only *Phialophora malorum* (Kidd and Beaum.) McColloch in addition to blue mold was found to have penetrated the fruit lenticels.

MATERIALS AND METHODS

Apples for the 1938 investigations were obtained from an orchard in the Wiley Heights district of the upper Yakima Valley of Washington. The fruit from this orchard had developed excessive amounts of lenticel infection in previous seasons. It was harvested at optimum maturity and sorted so as to be comparable with commercially packed fruit. Immediately or after various delay periods replicate samples were washed in a dual-process flood washer, first with sodium silicate (80 pounds to 100 gallons) at 100° F. and then with 1.5-percent hydrochloric acid at 80°. The period in each solution was 30 seconds. After washing, the fruit was dipped in a water suspension of blue mold spores, packed wet or dried overnight before packing, and stored at 32°.

The procedure was changed during 1939 and 1940 so that, instead of replicate samples from a single orchard, the fruit was obtained from five different orchards in the upper Yakima Valley. Handling methods were essentially the same as those employed in 1938 with the exception that all fruit was packed within a few hours after washing or inoculating. Also, the severity of the washing treatment was varied to determine its effect on the susceptibility of the fruit to infection. To ascertain the effect of a heavily contaminated washing solution on the development of decay, several rotten apples bearing large quantities of blue mold spores were added to the acid tank in one of the treatments and omitted in another; in these treatments the fruit was not subjected to inoculation subsequent to washing.

The 1941 work was undertaken to determine whether results comparable with those obtained in the previous three seasons could be obtained with naturally inoculated apples handled under commercial conditions. The Delicious apples used in this experiment were obtained from an orchard near Brewster, Wash., the fruit from which had shown serious amounts of lenticel decay over a period of years. The apples were fully mature when harvested and a few showed slight water core. A flood washer containing sodium silicate and hydrochloric acid was employed to clean the fruit. The apples were wrapped with oiled wrappers, packed in standard boxes, and held at 31° F. at a relative humidity of 85 to 87 percent.

The inoculum used in 1938 consisted of blue mold spores washed from the surface of naturally infected apples. During the succeeding 2 years a pure culture of *Penicillium expansum* was employed, the spores being obtained from inoculated fruit or from petri-dish cultures.

The details of other methods employed in some of the tests are presented in conjunction with the results under the appropriate headings.

FACTORS RELATED TO BLUE MOLD INFECTION

LENTICELS

In order to understand better the role of lenticels in blue mold infection, after different treatments in 1939 and 1941 epidermal tissues were stained by immersing the fruit in a solution of methylene blue. Clements (4),¹ whose method was followed in the present study, learned that after the treatment an open lenticel has a distinct halo of dye in the underlying parenchymatous tissue and that a closed lenticel remains uncolored or shows only a surface absorption of the dye. This method was used also by Baker and Heald (2) in studying factors affecting the incidence of lenticel infection by blue mold.

The details of the 1939 study are given in table 1. In only one lot (Lower Naches) did the number of lenticels on the pale and blush sides of the fruit differ significantly, and in only the Tieton apples were there significantly more open lenticels on one side than on the other. The lots did not differ significantly in the number of open or total lenticels on both sides of the fruit. The lot from Wiley Heights, which had the most lenticel invasions by blue mold, did not have the most open lenticels. (See table 6, p. 15.) Some factor related to the physiological condition of the lenticular tissue probably was responsible for the increased susceptibility of this lot.

The significantly greater number of open lenticels in the washed lots was correlated well with the higher percentage of lenticel infections found in the washed fruit. (See tables 4 and 5, pp. 9 and 10.)

¹ Italic numbers in parentheses refer to Literature Cited, p. 19.

TABLE 1.—Relation of handling treatments to the number of open lenticels of Delicious apples as indicated by immersion in 1:20,000 methylene blue, 1939

[Fruit was washed in sodium silicate (80 pounds to 100 gallons) at 110° and then in 1.5-percent hydrochloric acid at 100°. The following day it was immersed in dye for 36 hours, during which the temperature was gradually lowered from 65° to 45°. All temperatures in °F.]

Orchard and time and type of handling	Mean lenti circle	cels per 2-inch e ¹ on—	Mean open lenticels per 2-inch circle ³ on—			
	Blush side ²	Pale side ²	Blush side ²	Pale side ²		
Selah Heights: Day after harvest: Unwashed Washed Tieton:	Number $\left. \begin{array}{c} Number \\ 74.8 \pm 8.32 \end{array} \right.$	Number 82. 2±10. 75	$ \left\{ \begin{array}{c} Number \\ 15, 25 \pm 1, 92 \\ 26, 10 \pm 2, 07 \end{array} \right. $	<i>Number</i> 16. 70±1. 94 26. 95±1. 98		
Day after harvest: Unwashed Washed	$} 75.8 \pm 7.10$	87.0± 4.22	$ \begin{cases} 14.25 \pm 1.50 \\ 23.15 \pm 4.18 \end{cases} $	12.80 ± 1.02 35.85 ± 3.30		
Lower Naches: Day after harvest: Unwashed Washed.	$\left. \right\}$ 103. 0±10. 55	72.4± 5.97	$\begin{cases} 13.60\pm1.60\\ 23.10\pm2.59 \end{cases}$	11.15 ± 1.29 34.25 ± 5.22		
Naches Heights, Day after harvest: Unwashed. Washed. Wiley Heights:	$\Big\}$ 102. 0 \pm 3. 92	90.7 \pm 6.04	$\begin{cases} 15.35 \pm 1.66 \\ 27.30 \pm 3.15 \end{cases}$	17.80 ± 1.77 37.00 ± 4.09		
Day after harvest: Unwashed Washed	75. 2±14. 41	80.8± 6.96	$\begin{cases} 18.25 \pm 1.79 \\ 28.05 \pm 2.77 \end{cases}$	$14.90{\pm}2.07$ $31.95{\pm}2.21$		
Unwashed Washed	}		$ \left\{ \begin{array}{c} 10.85 \pm 1.12 \\ 16.35 \pm 2.04 \end{array} \right. $	$\begin{array}{c} 10.\ 65{\pm}0.\ 98\\ 18.\ 55{\pm}1.\ 71 \end{array}$		
After 3 days at 32": Unwashed Washed	}		$\begin{cases} 7.45\pm1.19\\ 14.25\pm1.00 \end{cases}$	$6.20{\pm}0.74$ 13.45 ${\pm}0.99$		
Unwashed Washed	}		$\begin{cases} 7.60\pm0.87\\ 15.60\pm1.69 \end{cases}$	7.50 ± 0.90 14.30 ± 1.67		

¹5-fruit sample.

² Standard errors after \pm .

³ 20-fruit sample.

In the 1939 test, holding the fruit in the warehouse or at 32° F. before washing reduced the number of open lenticels, but in 1941 (table 2) cold-storage delays failed to result in fewer open lenticels. Clements (4) had previously reported that warm dry air was effective in closing apple lenticels but that cold-storage temperatures were ineffective.

TABLE 2.—Effect of delay in cold storage prior to washing on the condition of the lenticels of Delicious apples and on the susceptibility of the skin to washing injury as indicated by immersion in 1:20,000 methylene blue, 1941

[Average of 10-apple samples washed in sodium silicate (63 pounds to 100 gallons) at 110° and then in 1.0percent hydrochloric acid at 100°. Immediately after washing the fruit was immersed in dye for 36 hours, during which the temperature was gradually lowered from 65° to 45°. All temperatures in °F.]

Time of washing	Open lenticels per fruit ^{1 2}	Washing injuries per fruit ¹ ²	Open lenticels and washing injuries per caylx basin ²
At harvest. After 2 weeks at 32°-34°. After 6 weeks at 32°-34°.	Number 23. 1 ± 5.38 27. 2 ± 4.71 28. 0 ± 10.4	Number 42. $8\pm 8. 24$ 28. $5\pm 6. 08$ 6. $7\pm 1. 22$	Number 25. 9±2. 88 35. 2±8. 26 9. 6±2. 54

¹ Exclusive of the calyx basin.

² Standard error after \pm .

Holding the fruit 2 weeks in cold storage before washing failed to reduce the number of openings (open lenticels and minute washing injuries) in the calyx basin (table 2), but the size of the injuries was considerably reduced. A delay of 6 weeks, however, caused a marked



FIGURE 2.—Radial sections of open and closed lenticels of Delicious apples. \times 116. A, Closed lenticel in which closure is due to partial cuticularization of the lenticel basin and to the formation of a compact layer of suberized cells. B, Closed lenticel in which an impervious layer of suberized cork has been formed. C, Open lenticel in which the basin lacks both cuticle and a compact layer of suberized cells. Note also large intercellular spaces in the hypodermis. reduction in the number of apertures that were penetrated by methylene blue. Very little decay developed in the naturally inoculated fruit used in 1941 (see table 7) despite the presence of numerous open lenticels and washing injuries.

A histological study of the lenticels of apples immersed in methylene blue corroborated the results obtained by Clements (4) that closure generally is due to the formation of a continuous layer of cuticle or to a combination of cuticularized and suberized cells (fig. 2, A). Lenticels rarely were closed by the formation of a layer of cork (fig. 2, B). In open lenticels frequently only a small fissure extended through the layer of modified cells in the lenticel basin (fig. 2, C).

Lenticels in apples washed the day after harvest did not differ histologically from those in unwashed fruit or in fruit washed after various holding periods. The layer of cuticle on parts of the fruit that developed washing injury was somewhat thinner than on those that were resistant to this damage. If the thickness of the cuticle is related to washing injury, it is not surprising that less injury and fewer open lenticels were found in the fruit delayed in the warehouse prior to washing than in apples washed immediately.

The cuticle was not significantly thicker on unwashed apples than on those cleaned in a dual-process wash consisting of sodium silicate (80 pounds to 100 gallons) at 110° F. and 1.5-percent hydrochloric acid at 100°.

MISCELLANEOUS INFECTION COURTS

When the so-called lenticel infections that developed in the fruit of the 1939 season were studied microscopically, it was found that penetration of true lenticels (those having a radial arrangement of the subepidermal cells) had occurred in only 63 percent of the 216 lesions examined. Approximately 23 percent of the invasions were through apertures not having the origin of true lenticels, and the remaining 14 percent apparently represented penetration of the unbroken epidermis. Some of the nonlenticular openings appeared to be minute washing injuries, but others were small punctures that apparently were caused by the fruit striking sharp granular material, possibly orchard sand in the bottom of field boxes. Many of the lenticel infections were found in bruised areas, and, as Baker and Heald (2) concluded, it is probable that localized pressure may have ruptured the layer of modified cells in the lenticel basin and thus increased susceptibility.

The relative importance of various infection courts in the initiation of blue mold decay in Delicious apples is shown in table 3. When a relatively severe dual-process washing treatment was used, most of the infection occurred at lenticels and washing injuries. With milder treatments washing injuries were relatively unimportant, but lenticels continued to predominate as places of invasion. Mechanical injuries were the most common avenues of infection in unwashed apples, but stem and calyx infections were rare in both washed and unwashed fruit. It appears from this study that acid and dual-process washing treatments predispose lenticels to penetration by blue mold; silicate solutions, however, appear to have little effect on lenticel susceptibility.

In the naturally contaminated apples used in 1941 (see table 7) very little lenticel infection occurred and about 73 percent of the blue mold decay originated at mechanical injuries. This fact, together with the evidence that many lenticel infections occur in bruised areas

and in lenticels injured by the washing process, emphasizes the importance of careful handling and mild washing in the control of blue mold decay.

TABLE 3.—Infection courts for blue mold in washed and unwashed Delicious apples

[Fruit harvested Oct. 2, 1939, or Sept. 25, 1940, washed in sodium silicate (80 pounds to 100 gallons) and then in 1.5-percent hydrochloric acid, dipped in a suspension of blue mold spores, packed, and stored at 32° until Jan. 15. In each treatment composite 50-apple samples from each of 5 orchards were used. All temperatures in ° F.]

aborring		Infections per 50 fruits					
showing washing injury		Lenticel	Stem and calyx	Mechan- ical injury	Visible washing injury	Total	
Percent	Percent	Number	Number	Number	Number	Number	
0.0	7.2	1.8	0.4	2.2	0.0	4.4	
25.2	21.6	8.2	. 2	4.0	1.8	14.2	
76.8	30.8	13.0	. 4	3.2	9.8	26.4	
. 0	24.4	9.8	. 2	4.0	.0	14.0	
. 0	4.0	1.2	. 0	1.0	.0	2.2	
30.8	14.8	4.0	. 2	3.2	1.0	8.4	
8.4	7.2	2.8	. 4	.8	. 2	4.2	
. 0	11.2	1.4	. 2	4.2	.0	5.8	
20.0	21.6	10.2	1.0	2.4	1.0	14.6	
. 0	17.8	5.8	. 2	4.2	.0	10.2	
.0	11.4	4.4	. 2	2.6	.0	7.2	
	washing injury Percent 0.0 25.2 76.8 0 30.8 8.4 0 30.8 8.4 0 20.0 0 0 0 0 0 0	$\begin{array}{c c} \text{washing} \\ \text{injury} \\ \hline \\ \hline \\ \hline \\ \hline \\ Percent \\ 0 \\ 25, 2 \\ 25, 2 \\ 21, 6 \\ 76, 8 \\ 0 \\ 24, 4 \\ 0 \\ 0 \\ 24, 4 \\ 0 \\ 0 \\ 11, 2 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 0 \\ 11, 4 \\ 0 \\ 10 \\ 10 \\ 10 \\ 10 \\ 10 \\ 10 \\ 1$	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	$ \begin{array}{c c c c c c c c c c c c c c c c c c c $	

Holding Fruit in Orchard or Warehouse Prior to Washing

An increased awareness among shippers of the necessity for better fruit quality and the provision of larger cold-storage facilities in the production areas have resulted in a tendency to remove apples from the orchard to cold storage with less delay than was the case some years ago. That prompt storage of the fruit at low temperatures would result in less decay than delayed storage seemed evident from the early work of Ramsey, McKay, Markell, and Bird (21); also Plagge, Maney, and Pickett (20) stated that prompt storage at 32° to 36° F. tended to hold soft rot caused by Penicillium under control. On the other hand, Baker and Heald (2) reported that there appeared to be a tendency for fruit held in the orchard for 6 to 12 days to develop less decay than that stored immediately. This apparent discrepancy they attributed to the fact that lenticels of the fruit probably became less susceptible to invasion because of dehydration and cutinization during the delay. The histological studies by Clements (4) showed that these processes can effectively close a high percentage of the lenticels of the apple.

One of the writers working with chemically treated packaging materials reported ² that 865 Delicious apples washed and packed immediately after harvest had developed 194 lenticel infections when examined late in January, whereas 938 fruits washed and packed after 10 days in the warehouse had developed only 30 infections. Both lots had been dipped in a suspension of blue mold spores before being packed.

² RYALL, A. L. EXPERIMENTS WITH CERTAIN CHEMICALS IN PACKAGING MATE-RIALS FOR CONTROL OF MOLDS IN APPLES AND PEARS. Unpublished Progress Rpt. 9 pp. 1936.

The data for 1938 (table 4) indicate that in general there were fewer lenticel infections and less total decay in fruit held 3 days in the warehouse than in that stored immediately; the differences were much greater with washed than with unwashed fruit. Usually somewhat larger percentages of the fruit held 3 days than of that stored immediately were infected at points other than lenticels.

TABLE 4.—Effect of various washing and packing treatments on blue mold decay in Delicious apples harvested Oct. 6, 1938

[Fruit washed in a flood washer, first with sodium silicate (80 pounds to 100 gallons) at 100° and then with 1.5-percent hydrochloric acid at 80°, dipped in a water suspension of blue mold spores, packed in oiled wrappers, and stored at 32° until Mar. 15, 1939. In each treatment 200-fruit samples were used. All temperatures in °F.]

	Blue mold decay							
	When							
Time and type of handling	At len	ticels	Fruits in-	-	After 6 addition-			
	Mean in- fections per 50 fruits	Fruits infected	fected at other points	Fruits infected	at 65°			
Wet when packed:		The second						
Day after harvest:	Number	Percent	Percent	Percent	Percent			
Unwashed	3. 3	6.5	5.0	11.5	12.0			
Washed	30.5	39.0	5.0	44.0	45.5			
After 3 days in warehouse:								
Unwashed	3, 3	5.5	9.0	14.5	15.0			
Washed	7.8	11.0	7.5	18.5	20.0			
After 2 weeks at 32°:								
Unwashed	0.0	0.0	10.0	10.0	11. 5			
Washed	1.3	2.5	7.5	10.0	12.0			
After 10 weeks at 32°:								
Unwashed	1.0	2.0	3.5	5.5	7.0			
Washed	6.0	10.5	4.0	14.5	17. 5			
Dry when packed:								
Day after narvest:	0.5	14.0	6.0	20.0	91 5			
Unwashed	9.0	14.0	10.0	20.0	21.0			
After 2 dama in worehouse.	54.0	45.0	10.0	55.0	00.0			
Unweshed	2.0	6.0	7.5	12.5	14 5			
Washed	12.0	10.0	5.0	24.0	97.0			
After 2 weeks at 20°	12.0	15.0	. 0.0	24.0	21.0			
Unwashed	1 3	2.5	12.5	15.0	16.5			
Washed	0.8	1.5	12.0	10.5	11 0			
After 10 weeks at 32°:	0.0	1.0	0.0	10.0	11.0			
Unwashed	1.0	1.5	5.5	7.0	9.0			
Washed	6.8	11.0	3.5	14.5	15.5			
	0.0		0.0	1110	1010			

Again in 1939 and 1940 (table 5) there were fewer lenticel and total infections in fruit held 3 days in the warehouse than in comparable fruit stored immediately. A delay in the warehouse apparently increases the resistance of the fruit to blue mold infection by reducing the number of open lenticels (table 1) and by decreasing the susceptibility of the fruit to washing injury. These results are in accord with those obtained by Baker and Heald (2) with fruit delayed in the orchard prior to storage.

TABLE 5.- Effect of handling treatments on incidence of blue mold decay in composite 50-apple samples from each of 5 orchards

[Fruit harvested Oct. 2, 1939, or Sept. 25, 1940, and washed in sodium silicate (80 pounds to 100 gallons) and then in 1.5-percent hydrochloric acid. All fruit, except as indicated, dipped in suspension of blue mold spores, packed in oiled wrappers, and stored at 32°. All temperatures in ° F.]

]	Results o	on Jan. 18	5	Results or 1	on Mar. Mar. 14, 1	18, 1940, 1941
Time and type of handling and washing	Fruits treated	Fruits	Fruits	Mean ir per 50	fections fruits	Fruits	Mean in per 50	nfections fruits
		injury	decayed	Lenticel	Total	decayed	Lenticel	Total
1939								
Day after harvest:	Number	Percent	Percent	Number	Number	Percent	Number	Number
Unwashed Silicate (110°) + acid (100°)	250	0.0	7.2	1.8	4.4	8.8 49.6	2.0	5.4
Silicate (100°) +acid (100°) -	248	25. 2	21.6	8.6	14.6	28.1	9.2	17.8
Acid (80°)	250	.0	24.4	9.8	14.0	31.6	12.0	17.8
Silicate (80°)	250	30.8	4.0	1.2	2.2	6.8 22.0	1.6	3.8
Silicate (105°)	249	8.4	7.2	3.0	4.4	9.2	3.0	5.6
Silicate (110°)+contaminated acid								
(100°) (not subsequently moc-	248	79.6	2.8	.2	1.4	6.8	6	3 4
Silicate (110°) +clean acid (100°)		10.0	2.0		1.1	0.0		0.1
(not subsequently inoculated).	248	80.0	.8	.0	.8	3.6	.6	2.4
After 3 days in warehouse: Unwashed	250	0	3.2	.0	1.6	6.0	.4	3.0
Silicate (110°)+acid (100°)	246	51.6	14.0	4.2	9.8	22.0	4.6	13.8
Silicate (100°)+acid (80°)	250	10.8	10.0	1.8	5.2	14.0	2.2	7.4
Alter 3 days at 32°: Unwashed	250	. 0	4.8	.4	2.4	9.6	1.0	4.8
Silicate (110°)+acid (100°)	249	51.6	20.8	6.4	15.2	31.6	7.0	20.8
Washed day after harvest and in-								
Silicate (110°) + acid (100°)	250	85.2	32.0	9.8	24.8	54.4	11.2	37.2
Silicate (100°) + acid (80°)	250	48.0	9.6	4.0	8.0	16.0	4.6	11.4
After 2 weeks at 32°:	250	0	16.4	3.6	0.8	20.4	28	11.8
Silicate (110°)+acid (100°)	250	40.4	26.8	14.2	22.4	34.8	15.8	26.4
Silicate (100°)+acid (80°)	248	2.4	24.0	13.6	19.2	32.0	15.8	24 . 0
After 6 weeks at 32°:	940	0	19	0	6	5.6	1	
Silicate (110°)+acid (100°)	249	20.0	$9.6^{1.2}$	3.2	6.6	20.4	6.8	12.4
Silicate (100°)+acid (80°)	247	1.2	5.2	1.4	2.8	12.0	2.8	6.6
After 10 weeks at 32°:	250	0	1.2	0	6	8.0	6	4 9
Silicate (110°)+acid (100°)	250	13. 2	3.2	.0	2.6	16.8	7.0	11.8
Silicate (100°) + acid (80°)	250	8	.8	.0	. 8	17.2	4.0	10.4
1940								
Day after harvest:								
Unwashed	249	.0	11.2	1.4	5.8	12.8	1.6	6.6
Acid (80°)	230	20.0	17.8	5.8	10.2	20.0	6.6	10. 0
Silicate (80°)	246	.0	11.4	4.4	7.2	14.2	5.0	8.4
Silicate (100°) +acid (80°) (not incompared)	250	95.9	1 9	2	6	1.6	2	8
After 3 days in warehouse:	200	20.2	1.2			1.0		
Unwashed	249	.0	8.4	1.4	4.2	10.4	1.6	5.2
Silicate (100°) + acid (80°)	248	6.5	9.7	2.2	5.2	14.9	3.4	8.2
Silicate (80°)	249	.0	7.2	2.0	4.0	13.6	3.0	7.6
After 2 weeks at 32°:	940	0	5.9		94	10.4	14	5.0
Silicate (100°) + acid (80°)	249	.0	20.5	8.8	12.6	24.5	9.0.	14. 6
Acid (80°)	250	0	13.6	3.2	7.2	16.4	4.0	8.6
Silicate (80°)	250	.0	20.4	8.2	13.0	22.4	8.2	14.0
Unwashed	249	.0	2.8	.8	1.8	8.0	1.4	4.4
Silicate (100°)+acid (80°)	249	.0	6.4	2.2	3.6	16.4	5.4	9.2
Acid (80°) Silicate (80°)	250	.0	4.0	1.2	2.0	14.0	3.0	8.9
After 10 weeks at 32°:	200		0.0	0.0	1.0	10.0	0.0	0. 2
Unwashed	1 200	.0	1.0	.0	.5	10.5	2.5	6.2
A cid (80°)	1 200	- 10. 7	3.5	.0	1.9	22.3	8.0	13.5
Silicate (80°)	3 147	. 0	.7	. 0	. 3	7.6	2.3	4.9

Fruit from only 4 lots included.
 Value indicates some error in washing treatment, probably uncontrolled solution temperatures.
 Fruit from only 3 lots included.

COLD-STORING FRUIT PRIOR TO WASHING

Since 1930 there has been a trend in the Pacific Northwest toward holding unpacked apples in cold storage and thus extending the packing season over the fall and winter months. Various opinions have been published as to the effect of washing and packing apples subsequent to refrigeration on the development of decay. Robinson and Hartman (22) and Hartman, Robinson, and Zeller (13) recommended washing immediately after picking as best for both residue removal and decay control. Baker and Heald (2) reported that the cold storage of Winesap and Delicious apples for 3 and 5 weeks, respectively, prior to washing increased both the amount of lenticel infection and the total blue mold decay. However, in these tests the fruit was dipped in a spore suspension immediately after it was picked; it is possible that the results would have been entirely different with naturally contaminated fruit or with fruit inoculated at the time of packing. In a later experiment the same investigators (3) compared the susceptibility of early and prime-maturity Delicious and Winesap apples to lenticel infection when coated with a paste of decayed tissue at harvesttime and after various periods of cold storage. In three of the four lots used there was a marked reduction in the number of lenticel infections when the inoculations were made after storage periods of 60, 120, and 180 days as compared with fruit inoculated immediately after harvest. There was some indication that susceptibility to lenticel infection was increased by storage for 240 days.

Evidence was not obtained in any of the 3 years (tables 4 and 5) that holding apples loose in cold storage for 2, 6, or 10 weeks before washing and packing definitely increases susceptibility to blue mold. When fruit was stored for 6 weeks its resistance to infection was consistently increased. In 1938 lenticel infections were decidedly fewer on the fruit held in cold storage. In 1939 the number of lenticel infections was reduced by 6- and 10-week delays but not by a 2-week delay. The results in 1940 were inconsistent, but in general there were slightly more lenticel infections and total decay in the fruit immediately washed than in that stored before washing.

Injury to the fruit from the washing process was invariably less when the fruit was washed after various delays in cold storage than when it was washed immediately after harvest (table 5). Except in the case of the fruit held for 10 weeks in 1940, the reduction in the amount of injury was directly related to the length of the storage period before washing.

The cold storage of apples in field boxes prior to washing has certain accepted advantages, such as more rapid removal of field heat, prevention of excessive accumulation of picked fruit in the orchard or warehouse, and the distribution of packing over a longer period. This practice permits a smaller investment in packing equipment, a longer period of employment for the packing-house force, and an opportunity to determine the marketing possibilities for a particular season before additional investments are made. From the present study it appears that storing the Delicious variety at 32° F. for periods up to 10 weeks prior to packing has the additional advantage of decreasing washing injury and susceptibility to blue mold infection. Against the advantages of cold storage in field boxes the packer

Against the advantages of cold storage in field boxes the packer has to weigh the disadvantages of the increased cold-storage space needed for unsorted, loose apples, the increased cost of handling loose fruit, the cost of refrigerating culls, and the possibility that scald might develop on apples not protected by oiled paper.

KIND AND TEMPERATURE OF WASHING SOLUTIONS

Most experimentation (5, 7, 10, 11, 12, 16, 17, 25) has indicated that mild washing to remove spray residue does not increase subsequent decay by blue mold (2, 13, 15) and other fungi. On the contrary, however, Pentzer (19) and Streeter and Harman (27) found greater decay in washed McIntosh apples and Green (9) reported that of five varieties studied only Jonathan showed increased decay, chiefly blue mold, after washing. Fisher and Reeves (6) stated that washing with proper equipment and due attention to sanitation did not impair the market value or the keeping quality of apples.

Little work has been done on the comparative effects of acid, alkaline, and dual washings on the subsequent development of decay. Overley, Overholser, and St. John (18) reported in 1934 that in preliminary experiments apples washed in sodium silicate solution developed less rot than comparable fruit washed in hydrochloric acid. In 1938 Haller and others (11), however, found no significant difference between the effects of these solutions.

In 1938 all the apples were washed with the same solutions (table 4). In most lots the washed fruit had more lenticel infection and total decay than the unwashed; the infections at other points, however, did not seem to be definitely related to the washing treatment. In five of the eight possible comparisons a slightly larger percentage of the unwashed than of the washed fruit had infections that did not start at lenticels.

In 1939 and 1940 the apples were washed with different solutions at the temperatures given in table 5. The most severe treatment (silicate at 110° F. plus acid at 100°) caused serious injury in the form of small skin cracks largely in and around the calyx basin. The less severe dual-process treatment and the acid wash at 105° resulted in moderate injury, whereas very little damage was caused by silicate at this temperature. At 80° neither acid nor silicate was visibly harmful. The work of Smith, Ryall, and Cassil (26) indicated, however, that a temperature of at least 100° is necessary if the spray is heavy.

In both 1939 and 1940 more lenticel infection and total decay occurred in fruit cleaned by the dual-wash process and by hydrochloric acid than in comparable unwashed apples. Fruit washed with sodium silicate alone at 80° F. was not consistently more susceptible than unwashed fruit. In the single comparison of apples washed with sodium silicate at 105° and unwashed fruit there was slightly more decay at lenticels of the former. In general, the susceptibility of the fruit was directly related to the severity of the washing treatment. Even where visible skin cracks did not occur, the washing treatment was found to increase the susceptibility of the lenticels to infection.

Size of Spore Population

The early work of Heald, Neller, and Overley (15) showed that the amount of decay in punctured Jonathan apples varied directly with the number of blue mold spores in the washing solution. Later Baker and Heald (2) found also that lenticel infection in several varieties of apples was directly related to the spore load of the fruit. In the present investigation opportunity was afforded to study further the importance of spore population in the initiation of decay. The data in figure 3 indicate that one of the critical factors in the



FIGURE 3.—Effect of blue mold spore population on infections in Delicious apples from five orchards. Washing was done at harvest in a dual-process machine containing sodium silicate (80 pounds to 100 gallons) at 110° F. and 1.5-percent hydrochloric acid at 100° ; it was followed by a rinse of clean water and in some instances by an immersion in a spore suspension. All lots were packed immediately after treatment and stored at 32° until the following March.

development of decay is the spore load carried by the fruit at the time of packing. Washed, noninoculated apples developed fewer lenticel and total infections than unwashed, inoculated ones and the latter fewer than those washed and inoculated. Where an adequate final rinse of clean water was used, the addition of spores to the acid solution caused only slightly more decay than when uncontaminated solutions were used. In both 1939 and 1940, immersing the fruit in a suspension of spores before packing resulted in many times the infections found in noninoculated fruit.

The commercial significance of this test is that it emphasizes the importance of a thorough final rinse of clean water in the control of blue mold decay. Experiments (15) have shown that thousands of fungus spores per cubic centimeter may accumulate in the washing solutions and in rinse tanks in which the water is recirculated. If these spores are not rinsed from the fruit prior to packing, much decay may develop in the stored fruit.

PACKING WET OR DRY

A number of investigators (9, 10, 11, 13, 14, 15) have studied the effects of packing washed apples when wet or dry on the subsequent development of decay; with few exceptions they concluded that packing apples while wet did not cause an increase in decay. Heald, Neller, and Overley (15) found that commercially sound and mechanically injured fruit packed wet and held in common storage developed more decay than comparable fruit packed dry. Robinson and Hartman (22, p. 31) reported that "fruit treated late in the season, after more or less contamination had taken place, seemed to decay more readily when packed wet."

In 1938 part of the fruit was packed wet and part the day after treatment, when the fruit was practically free of surface water. No evidence was obtained (table 4) that blue mold decay was increased by the fruit being packed wet. The apples packed wet presumably became dry before the end of the minimum period necessary for the germination of blue mold spores—10 or more hours at 65° F. or room temperature (1, 8).

FRUIT VARIABILITY

Records kept by commercial storage plants indicate that apples from certain orchards consistently develop more blue mold decay, particularly at lenticels, than those from other orchards. Baker and Heald (2) obtained similar data and also found that apples from other growers were severely decayed only in occasional years. They were unable to correlate susceptibility to lenticel infection with irrigation (2), fertilizer practice (2, 3), or fruit size (2), but they found that advanced maturity at harvest definitely predisposed the fruit to infection (2, 3).

In the present study it was found that fruit from the different orchards varied in susceptibility (table 6), the differences apparently being due both to seasonal factors and to conditions existing in some of the orchards. In 1939, apples from the Wiley Heights orchard developed more than twice as many lenticel infections as those from any other orchard, whereas in 1940 they developed the fewest infections. The data in table 1 show that the fruit from this orchard did

not have more open lenticels in 1939 than that from some of the other lots; so the large number of infections cannot be explained on that basis. The fruit from the Wiley Heights orchard was poorly shaped in both 1939 and 1940, but it was considerably larger in 1939. In 1939 these apples were highly susceptible to infection when dipped in a suspension of spores, but when they were not inoculated no decay developed (fig. 3). These results can be explained on the presumption that the natural spore load carried by the fruit was very light.

TABLE 6.—Relation of orchard source to the susceptibility of Delicious apples inoculated with blue mold

[Fruit harvested Oct. 2, 1939, and Sept. 25, 1940; total decay for March 18, 1940, and March 14, 1941, respec-tively. All fruit, except as indicated, dipped in a spore suspension, packed, and stored at 32° immediately after washing. All temperatures in ° F.]

	Infections ¹ in fruit from indicated orchard									
Time of washing	Tieton		Lower Naches		Naches Heights		Selah Heights		Wiley Heights	
	Len- ticel	Other	Len- ticel	Other	Len- ticel	Other	Len- ticel	Other	Len- ticel	Other
1939 ²	Num- ber	Num- ber	Num- ber	Num-	Num- ber	Num- ber	Num- ber	Num-	Num-	Num-
Day after harvest	15	28	19	26	14	34	22	37	54	46
Day after harvest 3	12	44	8	34	18	39	16	41	43	45
After 3 days in warehouse	1	11	9	11	5	22	6	14	15	26
After 2 weeks at 32°	6	13	23	19	38	35	35	27	75	38
After 6 weeks at 32°	0	4	10	8	12	17	9	10	19	20
After 10 weeks at 32°	0	7	12	10	16	24	8	12	22	20
Mean	5.7	17.8	13.5	18.0	17.2	28.5	16.0	23.5	38.0	32.5
1940 *										
Day after harvest	8	10	25	15	26	19	26	20	10	10
After 3 days in warehouse	8	12	14	11	- 8	14	16	14	2	10
After 2 weeks at 32°	5	10	27	19	26	17	24	14	$\tilde{2}$	14
After 6 weeks at 32°	3	1	21	14	17	17	16	12	1	5
Mean	6.0	8.3	21.8	14.8	19.3	16.8	20.5	15.0	3.8	9.8

¹ Total found in 150 fruits; in 1939, 3 lots of 50 fruits each were used; in 1940, 4 lots of 50 fruits each were

¹ Focal found in 150 truits; in 1959, 5 lots of 30 truits each were used, and the results were reduced to a 150-fruit basis.
 ² From each orchard are included 1 unwashed lot, 1 lot washed first with sodium silicate (80 pounds to 100 gallons) at 100° F. and then with 1.5-percent hydrochloric acid at 80°, and 1 lot washed first with sodium silicate solution at 110° and then with hydrochloric acid at 100°.
 ³ Held at 32° for 2 weeks before being inoculated and packed.
 ⁴ From each orchard are included 1 unwashed lot 1 lot washed mith codium silicate (80 pounds to 100 gallons).

⁴ From each orchard are included 1 unwashed lot, 1 lot washed with sodium silicate (80 pounds to 100 gal-lons) at 80°, 1 lot washed with 1.5-percent hydrochloric acid at 80°, and 1 lot washed first with sodium silicate at 100° and then with hydrochloric acid at 80°.

The fruit from the Lower Naches, Naches Heights, and Selah Heights orchards developed a moderate amount of lenticel infection during both seasons, whereas the Tieton apples consistently showed a high degree of resistance. The apples from all of these ranches were fairly well shaped, but the fruit from the Tieton orchard tended to be somewhat smaller than that from the others. Although Baker and Heald (2) were unable to correlate fruit size with susceptibility to lenticel infection, the work of Rose (23) and the experience of commercial fruit handlers indicate that large apples of a given variety generally develop more decay than small ones. The increased susceptibility of large fruit may possibly be due to the fact that its greater weight and coarser texture make it more subject to bruising and abrasion. Although maturity has a pronounced effect on lenticel infection (2, 3), it apparently was not a factor in this experiment as all of the fruit was harvested on the same day from orchards within a 5-mile radius and at approximately the same altitude.

DEVELOPMENT AND TYPES OF DECAY

The results of the test conducted in 1941 with relatively large quantities of naturally contaminated apples handled, washed, and packed under commercial conditions are shown in table 7. Although this fruit developed very little decay, the results corroborate those obtained in previous years with artificially inoculated apples. Only 48 (0.59 percent) of the 8,099 fruits used in the experiment showed decay after storage for 3½ months. Of the apples washed immediately 0.83 percent developed decay, while of those held 2 and 6 weeks in cold storage before packing 0.51 and 0.43 percent, respectively, did so. Of the fruits held in cold storage before washing, none showed decay at time of washing after they had been stored for 2 weeks, and only 2 infected fruits were found at time of washing after they had been stored for 6 weeks. Because of mechanical injuries these 2 apples probably would have been sorted out regardless of the decay.

Only two lenticel infections developed in the fruit that was packed, and these occurred in the fruit delayed for 6 weeks before washing. Blue mold decay was slightly more prevalent than gray mold rot caused by *Botrytis cinerea* Fr., and the decay caused by *Phialophora malorum* was third in importance. Most of the blue mold infections were at small punctures, whereas the gray mold centered at the calyx. It is possible that some of the gray mold infections were at washing injuries in the calyx basin; however, in some instances the fungus appeared to have attacked the sepals first and then to have advanced into the tissue of the fruit. Inasmuch as the apples had sufficient open lenticels and washing injuries to permit considerable infection, the small amount of blue mold decay probably was due to the low population of spores presumably carried by the fruit at the time of packing.

The amount of washing injury visible after storage was much greater on fruit washed at harvesttime than on that delayed for 2 or 6 weeks in cold storage before washing. Washing injury incurred after the 6-week delay was not appreciably different from that found on fruit stored for only 2 weeks prior to washing.

The relation of fruit size to decay can be summarized from table 7 as follows: In boxes containing 72 to 80 apples, 1.14 percent of the fruits were decayed; in those containing 88 to 100 apples, 0.65 percent; and in those containing 113 to 125 apples, 0.39 percent. These results substantiate observations made during the 2 preceding years that larger fruit is more susceptible to infection than smaller fruit.

SPORE POPULATION OF WASHING SOLUTIONS

To determine the spore population in different compartments of the washing equipment used in 1941, dilution cultures were prepared from samples of solution taken from the various tanks. As shown in table 8 the acid and alkaline washing solutions had very low concentrations of fungus spores and the rinse tank, although fairly heavily contaminated, contained relatively few blue mold spores. A study of the colonies that developed in the dilution plates revealed that most of the spores belonged to the following genera: *Dematium, Trichoderma, Phoma, Cladosporium, and Phialophora.* Only the last two genera contain important apple-decaying species. The gray mold fungus, *Botrytis cinerea*, was found only rarely.

[Fruit harvested Sept. 22, 1941, washed in sodium silicate (63 pounds to 100 gallons) at 110° F. and in 1-percent hydrochloric acid at 100°, and examined Jan. 7, 1942] TABLE 7.—Decay of Delicious apples commercially washed and packed at harvest and after storage at 32° to 34° F.

Fruits show-	ing ing injury	P = 2 = 2 = 2 = 2 = 2 = 2 = 2 = 2 = 2 =
ther infections	Causal fungus	Botrytis cinerea do do Neodo Neodo Botrytis cinera do. Aduemata sp. Botrytis cinera Phidophora malorum. Phidophora malorum. Phidophora malorum.
U	Location	Calyx Calyx (Calyx (Calyx (Puncture Oalyx Puncture Calyx Worrhhole Worrhhole Calyy
	Total	N_{ber}^{Num-} ber 10 10 10 22 22 22 11 11 11 11 11 11 11 11 11 11
ctions	Me- chani- cal in- juries	Num- ber 0 2 2 2 2 2 2 2 2 2 2 2 0 0 0 0 0 0 0
nold infe at—	Stems and ca- lyxes	Num-ber 0 ber 0 ber 0 ber 0 ber 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0 0
Blue 1	Lenti- cels	N_{ber}^{Num}
: P	Fruits de- cayed	Num- ber 1 1 1 1 1 2 2 2 2 2 3 3 1 1 1 1 1 1 1 1
	Boxes	Num- ber 06 10 10 10 10 22 22 23 33 11 11 11 11
Fruits per box		$\begin{smallmatrix} Num \\ ber \\ ber \\ ber \\ 100 \\ 113 \\ 125 \\ 100 \\ 113 \\ 125 \\ 100 \\ 113 \\ 88 \\ 88 \\ 88 \\ 100 \\ 110 \\ 113 \\ 125 \\ 110 \\ 110 \\ 113 \\ 125 \\ 110 \\ 125 \\ 110 \\ 125 \\$
Time of washing and packing		At harvest

¹ 9 infections at this point.

The spore-population samples were taken at the time of the experimental fruit washing, which was approximately 2 hours after the close of the day's commercial washing. The low population of viable spores in the tanks suggested the possibility that many spores had been killed by the chemical solutions in the interim between the commercial and the experimental washings. To obtain further information on this point, samples were taken during the course of commercial washing on the afternoon of November 1 and again 2 hours after commercial washing had ceased. The data (table 8) show that there were no outstanding differences between the commercial and the experimental washings with regard to spore populations. At the temperatures employed the washing solutions were evidently sufficiently lethal to prevent an appreciable accumulation of viable spores (1, 24). The low population of spores of blue mold and other apple-rotting fungi in the washing tanks undoubtedly was partly responsible for the low percentage of decay encountered in the 1941 experimental lots (table 7).

 TABLE 8.—Spore population in washing solutions and rinse water used on Delicious

 apples harvested Sept. 22, 1941

	Sam	ple A	Sample B		
Date, treatment, and washing solution	Total spores per cubic centi- meter	Blue mold spores per cubic centi- meter	Total spores per cubic centi- meter	Blue mold spores per cubic centi- meter	
Sept 22 (immediate treatment): Sodium silicate (63 pounds to 100 gallons) at 110°	Number 0	Number 0	Number 0	Number 0	
Hydrochloric acid (1-percent) at 100° Rinse water Oct. 6 (2-week delay treatment):	10 7, 000	$\begin{array}{c} 0\\ 20\end{array}$	0 5, 900	0 10	
Sodium silicate (63 pounds to 100 gallons) at 110° Hydrochloric acid (1-percent) at 100°	10	0	0	0	
Rinse water	5, 800	10	7, 700	10	
Sodium silicate (63 pounds to 100 gallons) at 110° Hydrochloric acid (1-percent) at 100°	80 0	$10 \\ 0$	90 10	10 0	
Rinse water Commercial wash: ¹	8, 200	10	5, 600	10	
Sodium silicate (63 pounds to 100 gallons) at 110°	210	40	140	10	
Hydrochioric acid (1-percent) at 100° Rinse water	5,100	0	0 4, 900	0 _ 10	

[All temperatures in ° F.]

¹ The samples for this determination were taken during the commercial operation of the washing machine, several hours prior to the experimental washing.

FUNGI ISOLATED FROM INFECTED FRUIT

No difficulty was encountered in distinguishing the lesions produced by *Penicillium expansum* (fig. 1) from those caused by other fungi. However, for verification a number of isolations were made from lesions classified as blue mold infections, and isolations were made from all infections which appeared to be caused by other pathogens.

The isolations from the lesions attributed to *Penicillium expansum* in all cases yielded this **species**. Isolations from lenticel infections which appeared different **from** blue mold yielded *Phialophora malorum* in all but one case; in this an unidentified species of *Penicillium* was obtained. The fungi isolated from lesions centered at other infection courts included the following in descending order of frequency: *Penicillium expansum*, *Botrytis cinerea*, *Phialophora malorum*, *Hormodendrum cladosporioides* (Fr.) Sacc., *Alternaria* sp., and *Neofabraea perennans*.

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